## Enzymax, LLC

# Product Name:RNA Gel Extraction KitCatalog No.:EZCR303Size:50 preps

### Product Information

Related products: RNA mini spin columns (30-50µl vol.) RNA Tini spin columns (5-20µl vol.) RNA Isolation kit (50) RNA Cleanup and Concentration kit (50) Catalog No. EZCR101 EZC107 EZCR300 702

- Easy and rapid with 20 min procedure using spin column

- High RNA recovery

- Extract and purify RNA fragments (70 bp-12 Kb) from standard or low-melting agarose gel in TAE or TBE buffer

#### **Kit Contents:**

Components	RNA Gel extraction kit 50 Preps (cat# EZCR303)
Gel Extraction Buffer	25 ml
3xWash Buffer*	8 ml
RNA Mini spin column with collection tube**	50

\*Add 100% ethanol before use: add 16 ml 100% ethanol to 8 ml 3xWash buffer.

\*\*Mini RNA Spin columns can be order separately for leftover solution (cat#EZCR101, \$59 for 100 columns)

#### **Caution:**

Gel Extracion Buffer contains chaotropic salt. Please use proper safety precautions and always wear gloves when handling the reagent. Avoid contact with skin, eyes or clothing. In case of accidental spill or contact, wash thoroughly with water, seek medical advice if necessary.

#### **Protocols:**

This kit is designed for RNA fragment recovery from agarose gel using Mini spin columns (Cat#EZCR101). If you have small amount of RNA sample on the gel, you can use Tini RNA Spin Columns (EZC107) with same protocol but cut down all the solution to 1/2 to 1/3. The elution volume for Tini RNA spin column can be as low as  $5\mu$ l.

- 1. Excise the RNA fragment from the gel with a clean scalpel, weight and transfer it to a clean tube. **Note:** Soak the gel in TE buffer for 30 min at room temperature if using formaldehyde agarose gel.
- Add 150 (3 gel volumes) or 300µl (6 gel volumes) Gel Extraction Buffer into each 50mg of gel slice. Note: Add 6 gel volumes of buffer if gel concentration is >2%.
- Incubate at 60°C for about 10 min till the gel slice is completely dissolved. Increase the temperature to 85°C, incubation time, or add more extraction buffer if the gel concentration is more than 2%. <u>Note:</u> If the color of the mixture turns a blue or purple color, adjust pH by adding a small volume (5µl) of 3M Sodium acetate (pH 5.0).
- 4. Add 1.5 volumes of 100% ethanol and mix by inverting the tube 3 times. **Note:** add equal volume of 75% ethanol for recovering RNA>200bp.
- 5. Load the sample mixture onto the **Mini RNA Spin Column** and spin in a microcentrifuge for 1 min at full speed (about 10,000 rpm). Do not load more than 700  $\mu$ l of sample on the Mini spin column at one time. Discard the flow through and load more sample mixture if needed.
- 6. Wash the column by adding 300 µl of **Wash Buffer** (ethanol added) and centrifuge for 1 min.
- 7. Wash once by adding 500  $\mu$ l of 80% ethanol and centrifuge for 1 min.
- 8. Discard flow through and place the column back in the same tube. Centrifuge the tube with lid open for 2 min. **Note:** this step is important since the residual ethanol may affect downstream applications)
- 9. Place the column in a clean 1.5 ml micro-centrifuge tube.
- 10. Add 50 μl or more **DNase/RNase water** or **TE** (10mM Tris/HCl, 1mM EDTA, pH8.0) (preheated at 65°C for better yield) to the **center** of the column and leave at room temperature for 5 min. Spin the column for 1 min to elute RNA from the column.

#### FOR RESEARCH USE ONLY

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